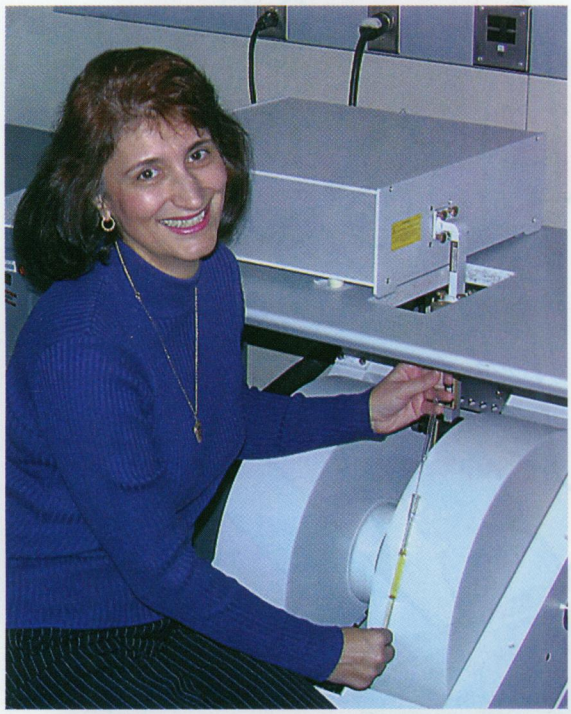


A stressful situation. Visiting scientist Maria Kadiiska is coordinating the evaluation of possible biomarkers of oxidative stress.

fying and detecting the presence of free radicals may be a significant step toward verifying the toxic components of air pollution particles, says Mason.

Another area the group has been studying is the production of nitric oxide (NO) in the metabolism of toxic chemicals and drugs. NO is an unusual free radical in that it is not highly reactive. It is commonly produced in metabolic processes and is usually beneficial, although overproduction of NO can have harmful effects.

The group has been studying the production of NO in the metabolism of the drug hydroxyurea, an anticancer agent that is also used to treat sickle cell anemia. In 1997, they determined that NO is produced when hydroxyurea is metabolized in rats, and they postulated that NO is also produced in humans who take the drug.

Recently, Richard Glover, a visiting fellow at the NIEHS, completed a study in humans with sickle cell anemia that was the first to indicate that hydroxyurea is metabolized to NO. Using ESR, the group detected nitrosyl hemoglobin within 30 minutes of oral administration of hydroxyurea to humans. Nitrosyl hemoglobin is NO bound to hemoglobin; thus is the presence of NO demonstrated. Because NO has been found to play a role in vasodilation (dilation of the arteries), the researchers suggest that the NO metabolite may be responsible for easing the pain of sickle cell anemia crises. This finding is significant both in that NO was directly detected in the blood of a human for the first time, and because it possibly sheds light on the role of NO in the drug's efficacy, says Mason.

Oxidative Stress Faculty

Markers of oxidative stress indicate the presence of free radicals in the body. It has been generally agreed among scientists who investigate free radicals that there is a need for validating sensitive and specific biomarkers for oxidative damage in rodents, nonhuman primates, and humans resulting from multiple types of oxidative insults. The NIEHS has taken the lead in coordinating the first comprehensive comparative study for determining which of the available biomarkers for oxidative stress are the most sensitive and selective. The study, called the Biomarkers of Oxidative Stress Study, was initiated by Carl Barrett, scientific director of the NIEHS, and is being organized and coordinated by Maria Kadiiska, a visiting scientist in the Free Radical Metabolism Section.

Researchers in 4 NIEHS laboratories and 12 outside laboratories are evaluating a battery of more than 25 assays as possible biomarkers of oxidative stress, using four models of oxidative stress: carbon tetrachloride poisoning, iron overload, ozone exposure, and paraquat poisoning. More than 2,500 samples of biological specimens are being evaluated. So far, Kadiiska says, two assays have emerged as the most promising for being truly indicative of oxidative stress—the measurement of isoprostanes and of protein carbonyls.

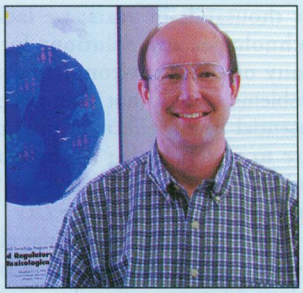
In principle, many free radicals can induce isoprostane generation. Isoprostanes are nonenzymatic prostaglandin-like products that result when free radicals catalyze the peroxidation of arachidonic acid. Their quantification appears to be a useful marker of oxidative stress such as the free radical oxidation of fats in the blood.

Protein oxidation products and carbonyl derivatives of proteins may result from oxidative modifications of amino acid side chains, reactive oxygen-mediated peptide cleavage, and reactions with lipid and carbohydrate oxidation products. The presence of carbonyl groups in proteins appears to indicate that the proteins have been subjected to oxidative free radical damage. Research has associated an increase in protein carbonyl content of tissues with several disorders, including rheumatoid arthritis, Alzheimer's disease, Parkinson's disease, and atherosclerosis.

The difficulty of measuring highly reactive free radicals by ESR or biomarkers in cell culture and humans is particularly challenging. But the importance of free radicals in biology, toxicology, and disease means many scientists will continue to pursue these goals.

Brandy E. Fisher

Stokes Recognized



William S. Stokes, associate director for animal and alternative resources at the NIEHS, has been recognized by The Humane Society of the United States under its Russell and Burch Awards Program. The program honors scientists who work to advance alternatives to the use of animals in research, testing, and education. Stokes was lauded for his ongoing work as director of the National Toxicology Program's Interagency Center for the Evaluation of Alternative Toxicological Methods, and as cochair of the Interagency Coordinating Committee on the Validation of Alternative Methods, which was established to review and implement alternatives to the use of animals in toxicity testing.

At the awards ceremony held 2 November 1998 in Washington, DC, Stokes received a framed certificate of recognition and a signed copy of the 1959 book *The Principles of Human Experimental Technique* by William Russell and Rex Burch. This book first introduced the concepts of replacement, reduction, and refinement, also known as the three Rs of laboratory animal welfare.